Exhibit 3

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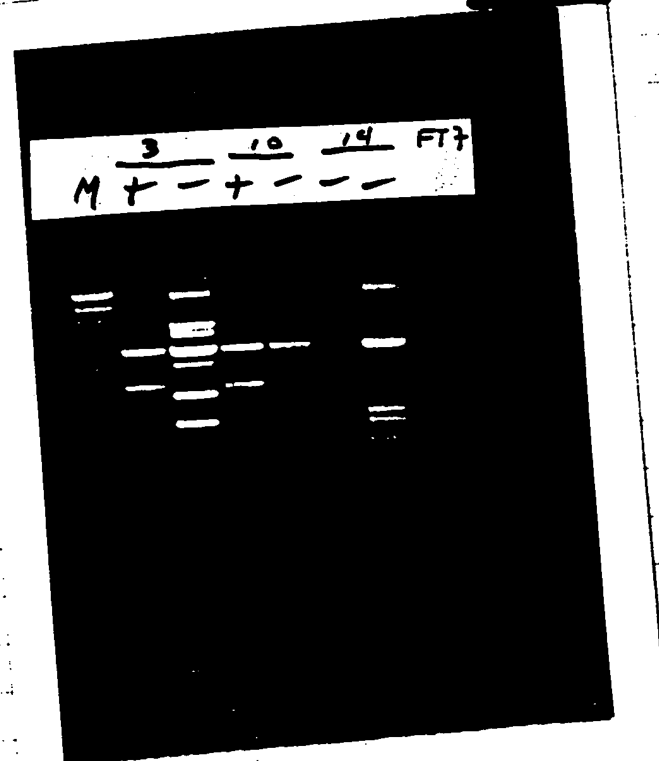
7/22 Digest FT7 cDNA 3, 10, 14 w/ KpnI/HindIII
use previous prep as control (-)

Expected Size

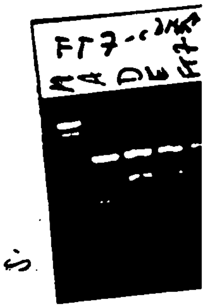
	(3) Sense	(4) Antis. (-)
3	3159	3159
	1573	1366
	625	782
10	3159	3159
	1573	1121
	650	832
	380	650
14	3159	3159
	1573	945
	650	832
	204	650



Major problem w/ #14

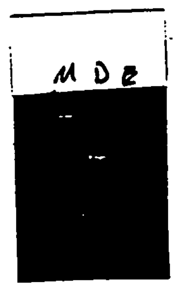
Some how samples got mixed-up

Go Bend & check out – Streak for single colonies
A B & E
Also mini prep from original culture
Digest w/ KpnI/Hind III O/N



7/23 Run gel of Digests
Clearly Sample A which was grown as maxi prep is
in the wrong orientation
Start O/N of D & E to mini prep before
starting 500ml culture

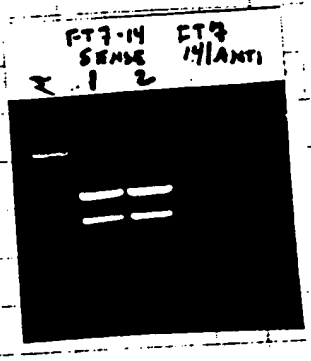
7/24 Mini prep FT7 cDNA 14-D & E
Digest w/ KpnI/HindIII
run on gel – Both are fine
use D for maxi



7/25 Plasmid isolation of FT7, cDNA 14 - D up to Banding
- do 500ml in 2 250 sets, end w/ 2 tubes to Band

7/26 Pull Bands - double band / prep - 6 hrs during the Day

7/27 Digest FT7.cDNA14 w/ KpNI / HindIII
① - Single band
② Double Banding
③ FT7-cDNA14 antisense



Check Absorbance / Concentration of Samples

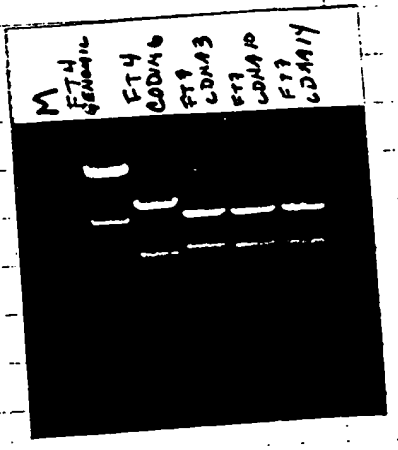
	260 FT7 cDNA 14	280		260/280
①	.127	.062	.63 ng/μl	2.0
②	.143	.065	.71 ng/μ	2.3

Sequence
FT7, cDNA3
T7
cDNA-14
T7
8850
9007
9874

Digest Mouse FT4

	260	280	260/280
PstI genomic	.085	.045	
NotI/XbaI Coding	.106	.060	

Digest of cDNA3
KpnI cDNA10
HindIII cDNA14



8/4 FACS Analysis of Transfected cells w/ following Vectors

2 plates/Vector — Divide

pcDNA I
FI7, 1, 2a 2b, 3
1, 2b, 3
1, 3
cDNA 3
cDNA 10
cDNA 14

FACS 7 mls
Yako 5 mls
EAT 3 mls (75 ul)
ETassy 5 mls (100 ul)

2nd Antibody
Fgm 2.5 mls
12.5/2.5 mls
Ig G 1.5
6.0/1.5 mls

Antibodies
IgM H — blue 1:100
IgM hf green 1:1000
IgM slx 1:200
IgG hc red 1:500
IgG slc blue 1:500

Results are
H - all neg
hf - all neg
slf - pcD (-), 1, 2a 2b, 3 (+), 1, 2b, 3 (+), 1, 3 (-), cDNA 3 (-), 10 (+), 14 (
hc all neg
slc all neg

8/8 Squam 12 pmm KG, FT4

6451	6080
2470	6199
6374	6087
6306	6086
6203	6085
5721	5671

8/9 Run sq gel of above samples
Also squam

5728	6084	7213
5731	5732	6082
5662	5727	6201
	5725	6200

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~~8/10 Sequencing gel of 8/9 samples~~
~~Sequence~~

FT4	6079	cDNA 14
	6202	T7
	6307	8850
	6373	8807
cDNA 10	715	8874
	946	

Protein assay of FACS Samples, Also CAT assay

pcDNA
FT7 1,2a2b,3
FT7 1,2b,3
FT7 1,3
cDNA3
cDNA10
cDNA14

BSA	Blank Protein		x̄
0	.100	.116	.108
1	.204	.225	.214
2	.369	.383	.376
4	.691	.673	.682
8	1.215	1.230	1.222
16			

5µl/Sample			x̄
pcDNA	.292	.294	.293
1,2a2b,3	.337	.330	.33
1,2b,3	.298	.343	.32
1,3	.369	.372	.37
cDNA3	.363	.379	.3
cDNA10	.306	.298	.3
cDNA14	.225	.253	.

FACS Results
Only stain w/ shx

1,2a2b,3	23.6%
1,2b,3	24.6%
cDNA10	14.9%
cDNA 14	8.0%

8/11 Sequencing of FT4 9/10 Samples

Micro BCA Protein Assay [redacted]

Reagent mix	MC	MB	MA
Per assay tube (ml)	0.01	0.24	0.25
Cocktail for Tubes			

Incubate 1 h at 60¡C and cool to room temp.
Since the color development has no end point, all tubes must be heated and cooled at the same time

1 mg/ml BSA (l)	Water (l)	Reagent (l)	Abs. 562	
0.0	500.0	500.0	Blank	Slope = 0.0734
1.0	499.0	500.0	0.108	Y intercept = 0.0656
2.0	498.0	500.0	0.214	X intercept = -0.8940
4.0	496.0	500.0	0.376	R = 0.9985
8.0	492.0	500.0	0.682	
16.0	484.0	500.0	1.222	

Sample	l in assay	Water (l)	Reagent (l)	Abs. 562	mg protein/ml
pcDNAI	5.00	495.00	500	0.293	0.62
FT7 1,2a2b,3	5.00	495.00	500	0.333	0.73
FT7 1,2b,3	5.00	495.00	500	0.320	0.69
FT7 1,3	5.00	495.00	500	0.370	0.83
cDNA 3	5.00	495.00	500	0.371	0.83
cDNA 10	5.00	495.00	500	0.302	0.64
cDNA 14	5.00	495.00	500	0.239	0.47

Cocktail 15
3H Chlor 300
Tris, 2M pH 8.0 75
But CoA 75
H_2O 300
50/tube

9/11 CAT Assay (FT7) samples
2.5 µl of cell extract
Control
pcDNA
1,2a2b,3
1,2b,3
1,3
cDNA 3
cDNA 10
cDNA 14

CAT [redacted]

Vector		Counts/5ul 8/12/94		Incorporated Counts (.95)		Total Counts		Total Counts Incorporated	
pcDNAI		11,349	11,829	9,189	8,063	236,169	244,643	9,673	8,487
FT7 1,2a2b,3		11,161	11,441	27,211	21,919	250,431	250,739	28,643	23,073
FT7 1,2b,3		11,772	11,826	37,541	40,684	272,981	277,204	39,517	42,825
FT7 1,3		11,215	11,690	23,076	28,706	247,376	262,506	24,291	30,217
cDNA 3		11,834	11,206	33,885	39,096	270,565	263,216	35,668	41,154
cDNA 10		12,017	11,312	30,066	33,165	270,406	259,405	31,848	34,911
cDNA14		11,079	11,570	44,133	40,529	265,713	271,929	46,456	42,662
Control			10,354		424		207,504		446
	Protein Conc. (ug/2.5ul)	Total Counts Inc- Bkg		% INC/hr		% INC/hr/ug		Mean CAT Activity	
pcDNAI	1.55	9,249	8,063	3.92	3.30	2.53	2.13	2.33	
FT7 1,2a2b,3	1.83	28,219	22,649	11.27	9.03	6.17	4.95	5.56	
FT7 1,2b,3	1.72	39,093	42,401	14.32	15.30	8.33	8.89	8.61	
FT7 1,3	2.07	23,867	29,793	9.65	11.35	4.66	5.48	5.07	
cDNA 3	2.07	35,244	40,730	13.03	15.47	6.29	7.48	6.88	
cDNA 10	1.60	31,224	34,487	11.55	13.29	7.22	8.31	7.76	
cDNA14	1.17	46,032	42,238	17.32	15.53	14.81	13.28	14.04	

9/12 Assemble Data of FT7 FACS, CAT Assay / give to Jucko
Work on FT4 sequence

9/15 7 deaza sequencing on Troublesome FT4 samples

6451	6200
6374	6079
6306	6307
6203	1897
5721	1898
	1899

9/16 Sequencing gel of 8/15 samples (FT4) Formamide gel
Probe Thomas's northern blot w/ GAP probe
To check condition of RNA

9/18 The 7 deaza technique didn't resolve all of the compressions
Try a Terminal transferase technique
Run standard sequenase rxn, after extension reaction
Heat tubes (A, C, G, T) for 1.5 mins 100 °C
Hold on ice 10 min, Prepare TdT / dNTP cocktail
Add to tubes, 37 °C 30 min
Add 5 µl Stop